ACCUMULATION OF ANTHROPOGENIC AND NATURAL ORIGIN ORGANOHALOGEN COMPOUNDS IN THREE PORPOISE SPECIES

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Introduction

Cetaceans are known to have high bioaccumulative capacity for anthropogenic organohalogen pollutants such as polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs) [1-3]. PCBs and PBDEs can be metabolized by Phase I cytochrome P450 monooxygenase (CYP) enzymes and Phase II conjugation enzymes [4,5]. Resemblance of hydroxylated metabolites of these two compounds (OH-PCBs and OH-PBDEs) to thyroid hormones (T_4) allows them to competitively bind to thyroid hormone transport protein, transthyretin (TTR) [6-8]. These metabolites potentially exert toxicities such as endocrine-disruption, neurotoxicity, reproductive and developmental toxicities [9-12] etc. Recent studies have shown that OH-PBDEs have alternative origins other than metabolic formation; they have been identified as natural products formed by marine organisms such as marine algae[13,14] and cyanobacteria associated with marine sponges [15,16]. OH-PBDEs can also be formed by demethylation of methoxylated polybrominated diphenyl ethers (MeO-PBDEs) [17]. The present study investigated the accumulation features of OH-PCBs, OH-PBDEs and the parent compounds in the blood of harbor porpoises, Dall's porpoises and finless porpoises stranded or bycaught along the Japanese coastal waters and the North Pacific Ocean. Additionally, the origins of these contaminants and factors affecting their pattern of accumulation were compared.

Materials and Methods

The blood samples were collected from the heart or the blood vessels of 13 harbor porpoises (*Phocoena phocoena*), 12 finless porpoises (*Neophocaena phocaenoides*) and 9 Dall's porpoises (*Phocoenoides dalli*) stranded or bycaught along the Japanese coastal waters and the North Pacific Ocean during 2005-2010. Six Dall's porpoises caught in the 1980s were also analyzed to assess the spatial distribution of the contaminants. Samples were stored in the Environmental Specimen Bank (*es*-BANK: http://esbank-ehime.com/) at Ehime University, Japan, at –25 °C until analyses.

62 PCB (mono- to deca-), 52 OH-PCB (tri- to octa-), 42 PBDE (mono- to deca-), 24 OH-/MeO-PBDE (tri- to octa-) congeners were analyzed in this study. The extraction and cleanup methods for PCBs and OH-PCBs as well as for PBDEs, OH-/MeO-PBDEs were described elsewhere [18,19]. OH-PCBs and OH-PBDEs were determined as MeO-PCBs or MeO-PBDEs using high-resolution GC/MS.

Results and Discussion

Accumulation Features of OH-PCBs

Concentrations of PCBs were the highest in Dall's pospoise (39,000 pg/g wet wt), followed by finless porpoise (26,000 pg/g wet wt) and harbor porpoise (6,500 pg/g wet wt). Inter-species comparison of the OH-PCBs concentrations showed the similar trend as PCBs, in which Dall's porpoise had the highest concentration (61 pg/g wet wt), followed by finless porpoise (23 pg/g wet wt) and harbor porpoise (14 pg/g wet wt). PCBs and OH-PCBs concentrations had a positive correlation (Fig. 3A; r=0.67, $p<0.001$), indicating that the large percentages of OH-PCBs found in porpoise blood were metabolically formed, rather than being taken up from the environment.

Distinct OH-PCBs congener patterns were observed among the three porpoise species. These species-specific accumulation patterns are possibly due to the differences in their metabolic capacity for PCBs, such as the induction of CYP enzymes, substrate specificity of phase I and II enzymes and/or affinity of OH-PCB congeners to TTR. From the results, the levels and accumulation patterns of OH-PCBs can possibly be explained by two factors: exposure levels of PCBs and/or metabolic capacities for PCBs (activities of CYP enzymes).

Accumulation Features of OH-PBDEs

Concentrations of PBDEs were the dominant in Dall's porpoise (2,300 pg/g wet wt), followed by finless porpoise (120 pg/g wet wt) and harbor porpoise (100 pg/g wet wt), and the PBDEs level in Dall's porpoise was more than one order of magnitude higher than the other porpoise species ($p<0.05$). On the other hand, OH-PBDEs showed accumulation pattern different from PBDEs. Finless porpoise had the highest OH-PBDEs level (2,000 pg/g wet wt), and the concentration was significantly higher than that of Dall's porpoise (680 pg/g wet wt) and harbor porpoise (240 pg/g wet wt). While OH-PCBs levels were about three orders of magnitude lower than PCBs, and OH-PBDEs levels were comparable with PBDEs or even higher in finless porpoise ($p<0.05$) (Fig. 1, 2). Moreover, no correlation was found between PBDEs and OH-PBDEs concentrations (Fig. 3B). These results suggest the levels of OH-PBDEs are not dependent on the PBDEs level and also OH-PBDEs have alternative source(s) other than the metabolic formation from PBDEs.

Among the 24 OH-PBDE congeners analyzed, 6OH-BDE47 was the most abundant, detected in all the samples and accounted for more than 95% of the total OH-PBDEs concentrations. 6OH-BDE47 is reported to be a natural product [20] although it is a minor metabolic product of rats exposed to PBDEs [21,22]. 2′OH-BDE68 was the second dominant congener, comprising 1-3% of the total, which is also formed by marine organisms [20]. In Dall's porpoise, several other congeners including 4′OH-BDE49 were identified, which is reported to be metabolic products of PBDEs [21]. Since PBDE and PCBs level in Dall's porpoise was more than one order of magnitude higher than in the other two species, exposure to higher concentrations of PBDEs might have induced CYP enzymes to form OH-PBDEs. From these results, OH-PBDEs found in porpoise blood can be considered to have both natural and anthropogenic origins although natural compounds were dominant.

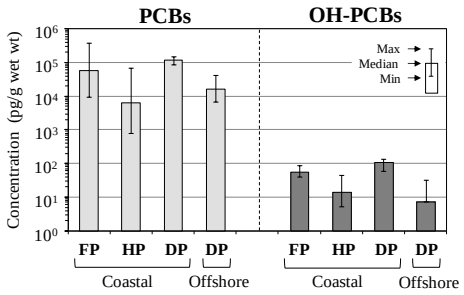

Fig. 1. Median concentrations of PCBs and OH-PCBs in the blood of finless porpoise (FP), harbor porpoise (HP) and Dall's porpoise (DP, coastal and offshore populations).

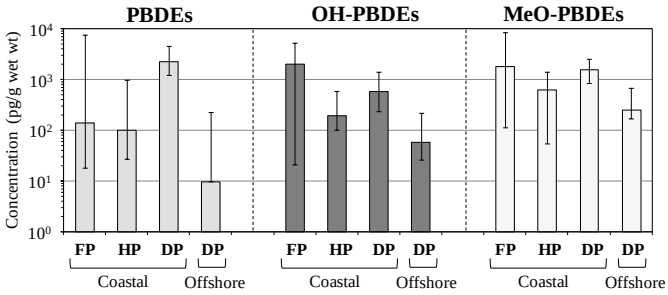

Fig. 2. Median concentrations of PBDEs, OH-PBDEs and MeO-PBDEs in the blood of finless porpoise (FP), harbor porpoise (HP) and Dall's porpoise (DP, coastal and offshore populations).

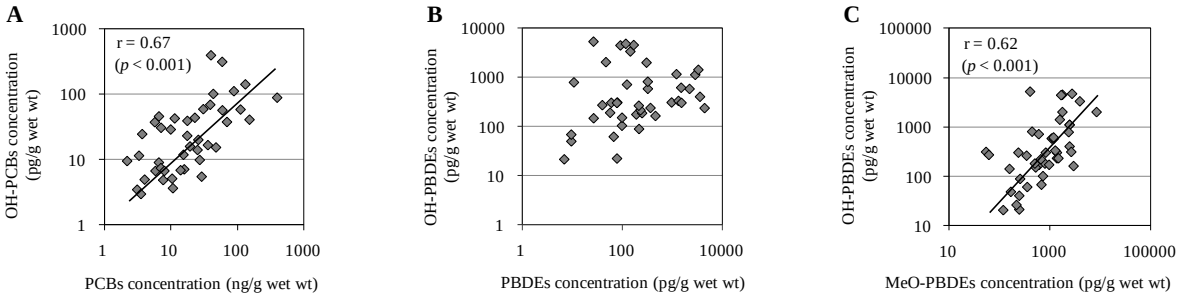

Fig. 3. Correlations between the concentrations of (A) PCBs (ng/g wet wt) and OH-PCBs (pg/g wet wt), (B) PBDEs (pg/g wet wt) and OH-PBDEs (pg/g wet wt) and (C) OH-PBDEs (pg/g wet wt) and MeO-PBDEs (pg/g wet wt) in the blood of three porpoise species.

Accumulation Features of MeO-PBDEs

MeO-PBDEs were found at comparable levels as OH-PBDEs, and the levels were the highest in finless porpoise (1,700 pg/g wet wt), followed by Dall's porpoise (1,600 pg/g wet wt) and harbor porpoise (650 pg/g wet wt). A strong positive correlation was observed between MeO-PBDEs and OH-PBDEs (Fig. 3C), which implies that these compounds share a common source such as the biosynthesis by marine organisms, or as a result of demethylation of MeO-PBDEs.

6MeO-BDE47 was the most abundant; accounting for more than 85% of the total concentrations, and 2′MeO-BDE68 was the second most abundant congener (6-15% total concentrations). These two congeners are analogous to the dominant OH-PBDE congeners, 6OH-BDE47 and 2′OH-BDE68, suggesting the formation of OH-PBDEs by demethylation. These two congeners were reported to be of natural origin by the analysis of radiocarbon studies of whale blubber specimens, and also detected from red algae and marine sponges [20,21,23].

Conclusion

OH-PCBs and OH-PBDEs have distinctive accumulation features, and the presence of natural origin OH-PBDEs make it impossible to predict their pattern of accumulation based on the patterns of OH-PCBs. It is necessary to further investigate the origin of OH- and MeO-PBDEs to understand their trends in accumulation in porpoise species as well as the other marine mammals. It is also important to investigate the potential toxicities of these natural-origin OH-PBDEs which accumulate in levels sometimes even higher than the levels of PBDEs.

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